Selectin ligands: 2,3,4-tri-*O*-acetyl-6-*O*-pivaloyl-α/β-galactopyranosyl halide as novel glycosyl donor for the synthesis of 3-*O*-sialyl or 3-*O*-sulfo Le^x and Le^a type structures

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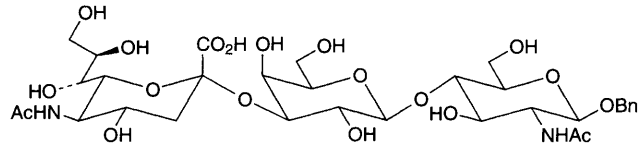
Stereoselective syntheses of 3-*O*-sialyl- and 3-*O*-sulfo- Lewis^x and Lewis^a type structures are accomplished through the use of key glycosyl donors 8 and 9.

The sialyl Lewis^x and sialyl Lewis^a structures are present in a wide variety of tumour-associated glycolipids and glycoproteins.[1] A number of investigators have reported increased levels of the sialyl dimeric Le^x antigen in metastatic tumours. Current research shows that sialyl Le^x and sialyl Le^a type structures act as ligands for selectins,[2] a family of membrane-bound cell adhesion molecules.[3] It is noteworthy that these selectins can also recognize the 3-*O*-sulfo Le^x and 3-*O*-sulfo Le^a structures.[4] All such observations have created an immense interest in the study and synthesis of sialyl Le^x, sialyl Le^a and the correspondent 3-*O*-sulfated moieties. Both chemical and biochemical approaches have been applied for the procurement of these compounds.[5] Recently, Danishefsky *et al.*[6] described an elegant synthesis of the sialyl Le^x compound. The interaction of these molecules with selectins suggests that such carbohydrate ligands can afford opportunities for the development of future drugs for the treatment of inflammatory diseases.



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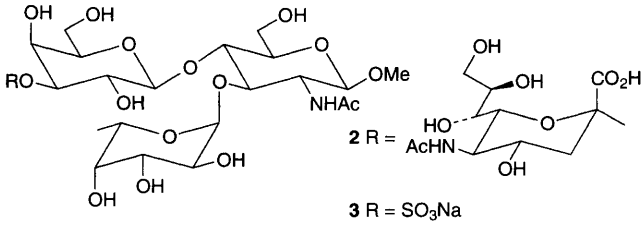
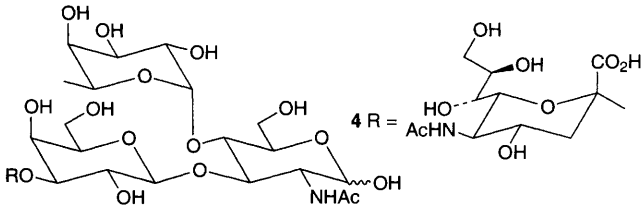


2 R =

3 R = SO_3Na

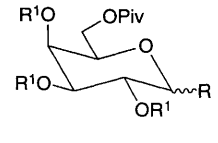


4 R =

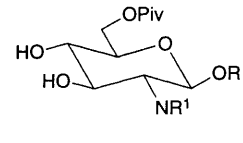
5 R = SO_3Na

Fig. 1 Target molecules: Sialyl lactosamine **1**; Sialyl and sulfated Le^x (**2** and **3**); Sialyl and sulfated Le^a (**4** and **5**)

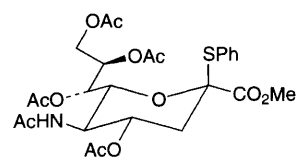
Advances made in the chemical synthesis of oligosaccharides suggest that glycosyl donors containing a permanent and a temporary protecting group are very important to the efficient synthesis of target compounds. Nicolaou *et al.*[5a] employed 2,4,6-tri-*O*-cetyl-3-*O*-chloroacetyl-β-D-galactopyranosyl fluoride for their synthesis of 3-*O*-sulfo Le^x type compounds. We hereby report that the title glycosylating reagents provide valuable donors for the synthesis of both 3-*O*-sialyl or 3-*O*-sulfo Le^x and Le^a type structures. Our strategy is based upon the observation that an *O*-acetyl group can be selectively removed in the presence of the 6-*O*-pivaloyl group to give 6-*O*-pivaloyl-β-D-galactopyranosyl-linked compounds which can then be selectively 3-*O*-sialylated or sulfated under appropriate conditions to yield the corresponding 3-*O*-sialylated or sulfated oligosaccharides. Compounds **1–5** (Fig. 1) were prepared from key intermediates **6–14**[7] (Fig. 2) by stereoselective transformation, as described in Schemes 1, 2 and 3, respectively. 1,2-3,4-di-*O*-isopropylidene-α-D-galactopyranose on treatment with pivaloyl chloride in pyridine and followed by hydrolysis with 70% aqueous acetic acid at 80 °C provided **6**, a mixture of α- and β-anomers in a ratio of 9 : 1, in 75% yield. *O*-Acetylation of **6** with pyridine–acetic anhydride, followed by treatment with 31% HBr–AcOH provided the mixture of α- and β-bromide **8** (9 : 1) in 90% yield. The bromide **8** was converted to its corresponding β-fluoride **9** by treatment with AgF in acetonitrile.[8] Glycosylation of **10** with **9** under Mukaiyama's conditions[9] ($SnCl_2$–AgOTf) afforded the β(1 → 3) linked disaccharide **15** in 17% yield and the β(1 → 4) linked disaccharide **16a** in 50% yield. De-*O*-acetylation of **16a** in MeOH–CH_2Cl_2



i **6** R = OH, R^1 = H
ii **7** R = OAc, R^1 = Ac
iii **8** R = Br, R^1 = Ac
9 R = F, R^1 = Ac



10 R = Bn, R^1 = Phth
11 R = Me, R^1 = Phth
12 R = Bn, R^1 = HAc



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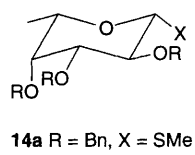
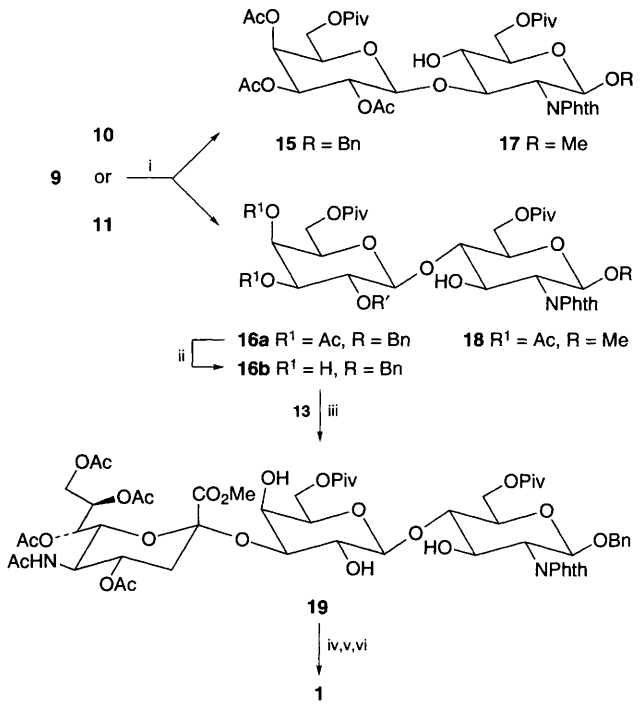
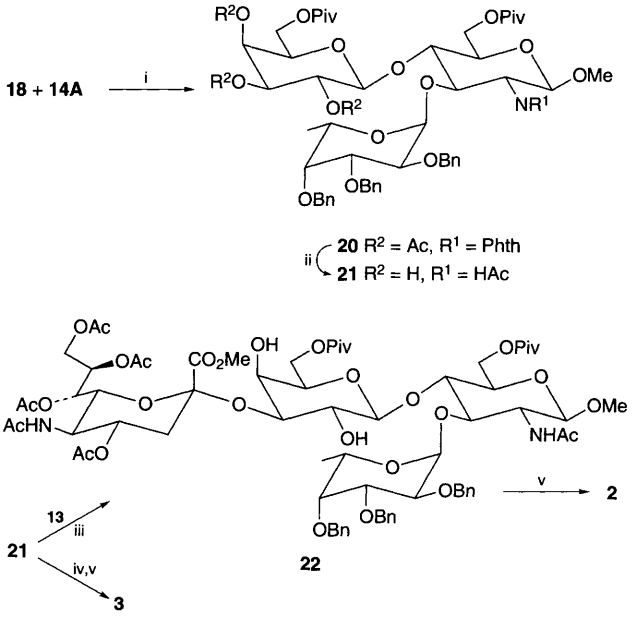


14a R = Bn, X = SMe
14b R = Bn, X = Br

Fig. 2 Key intermediates (**9–14**) involved in the synthesis of target compounds (**1–5**). *Reagents and conditions*: i, pyridine–Ac_2O (2 : 1, *v/v*), 16 h, 84%; ii, 31% HBr–AcOH, 16 h, 90%; iii, AgF–Acetonitrile, 16 h, 77%.



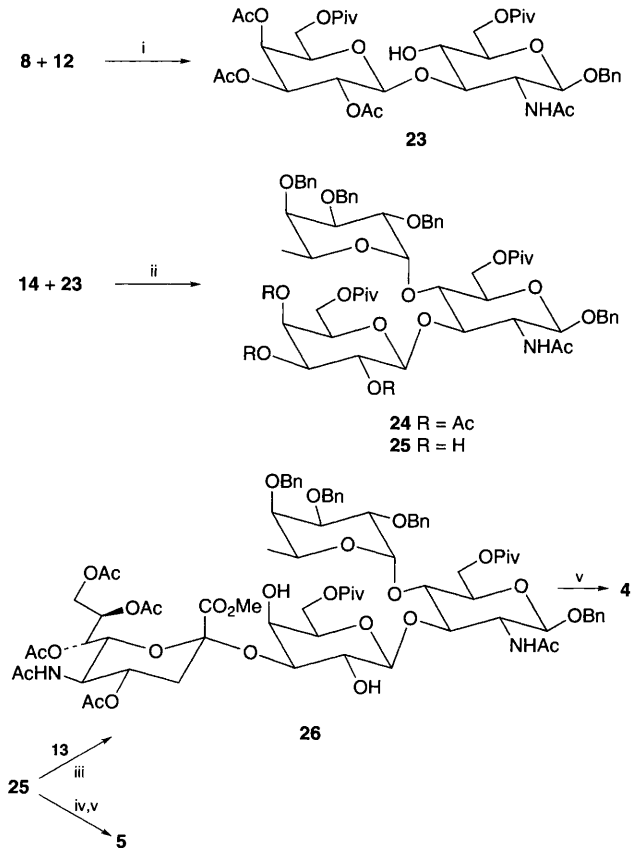
Scheme 1 *Reagents and conditions*: i, **9** (1.4 equiv.), AgOTf (1.2 equiv.), $SnCl_2$ (1.2 equiv.), 4 Å molecular sieves, CH_2Cl_2–toluene (5:1, *v/v*), −15 to 20 °C, 4 h, **15** (17%), **16a** (50%), **17** (21%), **18** (48%); ii, MeOH–CH_2Cl_2, 1:1 (*v/v*) (pH 10), 2 h, 0 °C, 78%; iii, **13** (2 equiv.), NIS (3 equiv.), triflic acid in propionitrile, −45 °C, 2 h, 53%; iv, LiI in pyridine (8 equiv.), 120 °C, 3 h, 75%; v, MeOH–hydrazine hydrate (5:1), *v/v*, 80 °C, 7 h, Ac_2O (excess), MeOH–CH_2Cl_2 (1:1, *v/v*), 0 °C, 1 h; vi, MeOH–MeONa, 48 h, 53% from **19**



Scheme 2 *Reagents and conditions*: i, **14b** (2 equiv.), **18** (1 equiv.), AgOTf (2 equiv.), 2,6-di-*tert*-butyl-4-methyl-pyridine (1.8 equiv.), 4 Å molecular sieves, CH_2Cl_2–toluene (2:3, *v/v*), −35 °C, 3 h, 79%; ii, EtOH–hydrazine hydrate (9:1, *v/v*), 100 °C, 6 h, MeOH–Et_3N–Ac_2O (4:2:1, *v/v*) 0 to 20 °C, 2 h, 62%; iii, **13** (2.5 equiv.), NIS–triflic acid in propionitrile (3 equiv.), −75 °C, 2 h, 66%; iv, SO_3–pyridine complex in pyridine (6 equiv.), 5 °C, 16 h; v, MeOH, 10% Pd–C, MeOH–MeONa, 72 h, H_2O, 5 h, **2** (96%), **3** (37% from **21**)

(1:1, *v/v*) with MeOH–MeONa (pH 10) at 0 °C provided the acceptor **16b** in 78% yield. Condensation with the sialic acid donor **13**[7e] under NIS–triflic acid catalysis[10] at −45 °C gave **19** in 53% yield. Similarly, formation of the 3,4-*O*-isopropylidene of **16b**, followed by α-L-fucopyranosylation with **14a** or **14b**, could be utilized for the synthesis of Le^y structures. Conversion of **19** into **1** was carried out systematically in four steps as outlined in Scheme 1.

The synthesis of **2** and **3** (Scheme 2) involved the glycosylation of **11** with fluoride **9** under conditions similar to those described for the preparation of **16a** (from **9**) to give the β(1 → 3) linked **17** and the β(1 → 4) linked **18** in 21% and 48% yields, respectively. The α-L-fucopyranosylation of **18** with **14a** under AgOTf–2,6-di-O-*tert*-butyl-4-methylpyridine conditions[11] furnished the fully protected trisaccharide **20** in 79% yield. Removal of both the phthalimido and acetate groups from **20** was accomplished by treatment with hydrazine hydrate in ethanol at 100 °C followed by *N*-acetylation to give **21** in 62% yield. Condensation of the sialic acid donor **13** with **21** under NIS–triflic acid condition at −75 °C[7a] provided **22** in 66% yield. The removal of *O*-benzyl (10% Pd/C), de-*O*-acetylation (MeOH–MeONa) and the addition of water to hydrolyse ester to acid afforded compound **2**. The selective sulfation of **21** with SO_3–pyridine complex in pyridine at 5 °C followed by the removal of protecting groups, as described for the preparation of **2** (from **22**), gave compound **3**.



Scheme 3 *Reagents and conditions*: i, **8** (1.5 equiv.), **12** (1.0 equiv.), $Hg(CN)_2$ (1.5 equiv.) in benzene–nitromethane (1:1, *v/v*), 55 °C, 16 h, 65%; ii, **23** (1.0 equiv.), **14a** (2.0 equiv.), $CuBr_2$ (3.0 equiv.), Bu_4NBr (3.0 equiv.), $ClCH_2CH_2Cl$–DMF (5:1, *v/v*), 4 Å molecular sieves, 16 h, 56%; iii, **13** (2.5 equiv.), NIS–triflic acid in propionitrile (3.0 equiv.), −75 °C, 2 h, 54%; iv, SO_3-pyridine complex in pyridine (6 equiv.), 5 °C, 16 h; v, MeOH–10% Pd–C, MeOH–MeONa, 7h, H_2O, 5 h, **4** (66%), **5** (50% from **25**)

The reaction of **12** with bromide **8** (Scheme 3) in benzene–nitromethane (1:1, *v/v*) at 55 °C afforded **23** in 65% yield. Similarly, **23** after de-*O*-acetylation, could be utilized for the preparation of Le^b structures as described for the preparation of Le^y structures from **19**. Glycosylation of **23** with **14b** under $CuBr_2$–Bu_4NBr[12] furnished trisaccharide **24** in 56% yield. The synthesis of **4** and **5** from **25** was achieved by a sequence of reactions similar to those described for the preparation of **2** and **3** from **21**. The structures of **1–5** were confirmed by ^1H and ^{13}C NMR and FAB mass spectroscopy.†

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Footnote

† ^1H and ^{13}C NMR spectra were recorded with a Bruker AM400 instrument at 400 MHz and 100.6 MHz respectively. *Selected data* for **1**: $[\alpha]_D$ −21 (*c* 0.5, H_2O); ^1H NMR (D_2O) δ 7.52–7.42 (5 H, m, arom.), 4.94 (d, *J* 12.2 Hz, H-1), 4.59 (d, *J* 8.1 Hz, H-1′), 2.81 (dd, *J* = 4.6 Hz, H-3″e), 2.08 and 1.97 (each s, 2 × NAc) and 1.84 (t, *J* 12.1 Hz, H-3″a); ^{13}C NMR δ 101.56 (C-1′), 98.85 (C-1), 98.81 (C-2″), 77.43 (C-3′), 77.29 (C-4), 61.59 (C-9″), 59.98 (C-6′), 59.01 (C-6), 54.02 (C-2), 50.68 (C-5″) and 38.63 (C-3″); *m/z* 765.3 $[M + H]^+$ and 786.8 $[M + Na]^+$. For **2**: $[\alpha]_D$ −38 (*c* 0.4, H_2O); ^1H NMR (D_2O) δ 5.09 (d, *J* 3.9 Hz, H-1″), 4.81 (d, *J* 7 Hz, H-1), 4.76 (d, *J* 7 Hz, H-1′), 3.50 (s, OMe), 2.76 (dd, *J* 4.6 Hz, H-3‴e), 2.03 and 2.02 (each s, 2 × NAc), 1.79 (t, *J* 12.1 Hz, H-3‴a) and 1.16 (d, *J* 6.6 Hz, H-6″); ^{13}C NMR δ 100.74 (C-1′), 100.65 (C-1), 98.67 (C-2‴), 97.56 (C-1″), 74.67 (C-3′), 74.30 (C-3), 73.88 (C-4), 61.61 (C-9‴), 60.44 (C-6′), 58.67 (C-6), 56.10 (OMe), 54.59 (C-2), 50.70 (C-5‴) and 14.24 (C-6″); *m/z* 833.3 $[M - Na]^-$. For **3**: $[\alpha]_D$ −45 (*c* 0.6, H_2O); ^1H NMR (D_2O) δ 5.15 (d, *J* 4.4 Hz, H-1″), 4.62 (d, *J* 7.8 Hz, H-1′), 3.54 (s, OMe), 2.07 (s, NAc) and 1.21 (d, *J* 6.6 Hz, H-6″); ^{13}C-NMR δ 100.72 (C-1′), 100.45 (C-1), 97.54 (C-1″), 79.20 (C-3′), 74.24 (C-3), 73.82 (C-4), 60.31 (C-6′), 58.70 (C-6), 56.11 (OMe), 54.62 (C-2) and 14.23 (C-6″); *m/z* 622.3 $[M - Na]^-$. For **4**: $[\alpha]_D$ −36 (*c* 0.8, H_2O); ^1H NMR (D_2O) δ 5.11 (1 H, d, *J* 3.0 Hz, H-1″), 4.56 (1 H, d, *J* 7.7 Hz, H-1), 4.52 (d, *J* 7.7 Hz, H-1′), 2.77 (dd, *J* 4.6 Hz, H-3‴e), 2.04 and 2.03 (6 H, each s, 2 × NAc), 1.76 (t, *J* 12.1 Hz, H-3‴a) and 1.17 (d, *J* 6.6 Hz, H-6″); ^{13}C NMR δ 101.77 (C-1′β), 98.39 (C-1′α), 98.35 (C-2‴), 96.99 (C-1″), 93.73 (C-1β), 89.96 (C-1α), 75.07 (C-3β), 74.64 (C-3′), 74.58 (C-3α), 73.71 (C-4β), 73.58 (C-4α), 61.27 (C-9‴), 60.61 (C-6′β), 60.58 (C-6′α), 58.76 (C-6β), 58.71 (C-6α), 55.85 (C-2β), 52.95 (C-2α), 50.67 (C-5‴), 39.02 (C-3‴) and 14.33 (C-6″); *m/z* 819.3 $[M - H]^-$. For **5**: $[\alpha]_D$ −41 (*c* 0.9, H_2O) [lit[5b] −38° (*c* 0.5, MeOH)]; ^1H NMR (D_2O) δ 5.06 (d, *J* 3 Hz, H-1″), 2.11 (s, NAc) and 1.22 (d, *J* 6.6 Hz, H-6″); ^{13}C NMR δ 101.59 (C-1′β), 99.44 (C-1′α), 97.01 (C-1″), 93.77 (C-1β), 89.94 (C-1α), 79.33 (C-3′), 79.22 (C-3β), 75.21 (C-3α), 74.58 (C-4β) and 73.50 (C-4α); *m/z* 608.3 $[M - Na]^-$.

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